

THE EFFECTS OF KINETIN AND ETHREL ON GERMINATION AND SEEDLING GROWTH OF CUCUMBER

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ABSTRACT

Effects of seed soaking with plant growth regulators (kinetin and Ethrel) on cucumber seedling emergence and seedling growth under normal conditions were studied to determine their usefulness in germination enhancement. The present study showed that seed treatment with ethrel and kinetin exhibited the inhibitory effect on seed germination and seedling growth. None of the seed treatments provided superior germination over the control.

KEYWORDS: Kinetin, Germination, Ethrel

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INTRODUCTION

Cucumber (*Cucumis sativus*) is a widely cultivated plant in the gourd family, Cucurbitaceae. It is a creeping vine that bears cylindrical fruits that are used as culinary vegetables. There are three main varieties of cucumber: *slicing*, *pickling*, and *burpless*. Within these varieties, several different cultivars have emerged. The cucumber is originally from Southern Asia, but now grows on most continents. Many different varieties are traded on the global market.

Seed germination is a complex physiological process that it is response to environmental signals such as water potential, light, nitrate and other factors. Some organic chemical substances of varied chemical affect seed germination, growth and development of plant. Organic chemical substances other than nutrients which are active in low concentration in promoting, inhibiting or otherwise modifying growth and development may be called growth regulators. Hormones of plants are organic substances in small amounts which regulate intracellular processes and change physiological and biochemical reactions in plants. The regulatory function of phytohormones in plants has been established by various workers (Amasino, 2005, Bandurski & Krekule, 1998, Becker & Hedrich, 2002, Fuchs & Lieberman, 1968, Kaminek *et al.*, 2006, Miller, 1961, Trewavas, 1991, Wang, 2001, Weyers & Paterson, 2001). Plant hormones have been studied for many years because they have been heavily implicated in the control of plant growth and development. They may regulate a wide variety of physiological responses in plants, are central in the regulation of elongation growth and are important in transduction of signals, respiration, water uptake, transport of ions in plant cells, phloem unloading, activation of proteinase inhibitor genes and gas exchange. Phytohormones have been considered to fall into six classes: auxins, cytokinins, gibberellins, abscisic acid, ethylene and brassinosteroids.

Cytokinins are a class of phytohormones that play an important role at all phases of plant development from seed germination to senescence (Mok & Mok, 1994, Niazi *et al.*, 2005, Riefler *et al.*, 2006). They act at the

cellular level by inducing expression of some genes, promotion mitosis and chloroplast development but also on the organ level by releasing buds from apical dominance or by inhibiting shoot and root growth (Osborne, 1962, Yaronskay *et al.*, 2007). Cytokinins have the unique ability to alleviate the effects of various inhibitors in seeds and other organs in a wide variety of plants. Kinetin, the most known cytokinin, has furfuryl ring at the N6-position of adenine and was identified in both animal cellular DNA and plant tissue extracts (Barciszewski *et al.*, 2000). Although its role for animals is well known, in the case of plants, it needs further investigation. Kinetin in low concentrations influences plants in a positive way but higher concentrations are toxic. Kinetin has been found to contain carbon, hydrogen, oxygen in the ratio C10 H9 N5 O. Kinetin has important functions in living organisms, in particular in animals and plants (Loneragan & Webb, 1993, Osborne, 1962). Kinetin is known to be essential to plants and is a necessary hormone for these organisms. Since their discovery cytokinins have been shown to have effects on many other physiological and developmental processes, including leaf senescence, nutrient mobilization, apical dominance, the formation and activity of shoot apical meristems, floral development, the breaking of bud dormancy and seed germination. Although certain external manifestations in plants in response to some of the chemicals are known today, yet in view of their differences in the behaviour in different plant species, it was proposed to study the effect of kinetin, adenine, uracil and thymine on the vegetative and reproductive growth of Cucumber.

Ethylene and ethrel, an ethylene-releasing compound, also release dormancy in various seeds (Esashi & Leopold, 1969, Ketrang & Morgan, 1969, Ketrang & Morgan, 1971, Tao *et al.*, 1974). Ethrel containing the active substance ethephon, is a plant growth regulator. Upon metabolism by the plant, it is converted into ethylene, Ethrel (2-chloroethanephosphonic acid) cause; effects similar to ethylene, and apparently breaks down in plant tissues releasing ethylene to a site of action. Ethylene is now well-known as an endogenous bio-regulant which actively affects almost all phases of plant growth and development including seed germination.

MATERIALS AND METHODS

The experiment was conducted in the department of crop physiology, Assam Agricultural University, Jorhat. The experiment was laid out in a completely randomized design (CRD) with three replications. For conducting the experiment cucumber seeds were collected from local market. Seeds were surface sterilised with 1.0% sodium hypochloride for five minutes and then washed with distilled water for three times. The petri dishes (9 cm) were also washed properly and oven dried before the experiment conduction. Predetermined concentrations of kinetin and ethrel solutions were prepared for the experiment (Table 1). To evaluate the effects of kinetin and ethrel, the seeds were placed in 9-cm Petri dishes (10 seeds per petri dish) on two layers of Whatman No. 1 filter paper moistened with 5 ml of distilled water or appropriate hormone solution (kinetin or ethrel). Radicle protrusion of 3 mm was scored as germination. Germination was counted in 24 hours intervals. The counting continued till seven consecutive days. Final germination percentage (%), germination index, vigour index, seedling length (cm), plumule length (cm), radicle length (cm), fresh and dry weight (g) of radical and plumule was recorded after 7 days of planting on filter paper. Germination percentage was determined by the following formula:

$$\text{Germination Percentage (GP)} = \frac{\text{no. of seeds germinated}}{\text{total no. of seeds}} \times 100$$

The caliper or scale was used to determine the length of radicle and plumule. Also, the samples were measured by digital scales to determine the fresh weight of radicle and plumule, as well as to determine the dry weight of radicle and plumule; the samples were placed in the oven for two days at 70 °C, and then measured by digital scales. The vigor index was calculated according to following formula,

Vigor Index (VI) = [seedling length (cm) × germination percentage]

Germination Index (GI) was figured as depicted in Association of authority Seed Analysts (1983) as the accompanying formulae:

Germination Index (GI) = number of seedlings emerged/number of days of first count + number of seedlings emerged/number of days of second count + + number of seedlings emerged/number of days of the last count.

The experiment was conducted with a total of 10 replicates per treatment and was repeated thrice. The percentage data were subjected to arcsine transformation. The data were recorded and analysed statistically in a completely randomised design by one-way analysis of variance (ANOVA) followed by Duncan's multiple-range test at a significance level of $P < 0.05$.

RESULTS

• Seed Viability and Seedling Vigour

Seed viability (germination percentage) and seedling vigour (lengths of hypocotyl, radical and entire seedling; as well as seedling fresh and dry weight) were studied in this experiment and results showed that there is no significant difference among the treatments and control for germination percentage. Germination index (24.90) was highest in control along with ethrel 50ppm (23.70) and ethrel 100 ppm (23.10). No significant difference among these three. Lowest germination index (5.90) was seen in kinetin 10 ppm (Table 2).

In case of radical, plumule and seedling length none of the treatments provided superior length over the control. Control having highest radical length (6.60 cm), plumule length (6.32 cm) and overall seedling length (12.92 cm) (Table 2). But, Ethrel 1 ppm also showed similar plumule length (6.61 cm) as control one. Probably the reason for reduction of root and shoot growth due to utilization kinetin and ethrel lead to reduction or lack of transfer of nutrients from cotyledon to embryo. The results of mean comparison of data show that there is a difference between the effects of different ethrel and kinetin levels on seedling fresh weight and seedling dry weight. So that the highest seedling fresh weight was observed in ethrel 1 ppm (3.192g) and highest seedling dry weight was observed in kinetin 50 ppm (0.1660 g) and lowest dry and fresh weight was reported in kinetin 100 ppm (0.0964 g) and ethrel 50 ppm (1.520 g) respectively (Table 2).

• Seed Vigour Index

Kinetin and ethrel applications decreases seed vigour index in cucumber as compared with the control (Table 2). Highest seed vigour index was recorded in control (1292) and lowest was recorded in kinetin 10ppm, kinetin 100 ppm, ethrel 50 ppm and ethrel 100ppm.

DISCUSSIONS

Under the experimental conditions used here, seed treatment with different concentration of Kinetin, ethrel has not shown any significant response in germination percentage as compared to the control. Kabar (1989) reported that experiments with various dicot seeds, treatments with kinetin were observed to promote total percentage germination of these seeds under saline conditions much more than GA_3 . But on contrary in this experiment, statistical analysis of the germination percentage, no significant difference among the mean was found. Whereas the germination index was

significantly higher in the control which was followed by Ethrel 50 ppm and Ethrel 100ppm treatment in the same significance range. The effect of kinetin in germination index was lowest in Kinetin 10 ppm. Lowest growth rate was reported by Majumdar & Boissya (1983) for kinetin treated cucumber which is evidently supported by this study. Thus the findings of the present investigation are contrary to the most of previous researches where kinetin was reported to be capable of overcoming the inhibition of germination. A negative response of kinetin stimulation in japonica rice cultivar was reported (Miyoshi & Sato, 1996) which is supported by the present findings in cucumber. In this study, light and temperature were not the parameters under study. Therefore further study on effect of kinetin and ethrel within specific temperature and light is suggested as Welch (1976) reported of Kinetin treatments to significantly improve germination of seeds lettuce varieties at the 25°C storage temperature. It was reported that PGRs (GA₃ and KIN) overcame the allelochemical stress on seed germination and seedling growth by affecting root and shoot growth rates in several species (Terzi & Kocaçalışkan, 2009). Decrease of cytokine indigenous levels in plants under stress refers to this possibility that reduction of cytokine limits growth in plants under stress and external application of kinetin could lead to increase seedling weight and seedling dried weight under stress (Hare *et al.*, 1997). Increase of seedling weight and seedling dried weight and shoot weight under stress by kinetin could be related to increase of water intake due to permissively of membrane and osmotic active minerals inner concentration (Stavir *et al.*, 1998). Decrease in activity in seeds causes to reduction of formation of glucose from starch and decreases in sucrose synthesis. This conditions lead to limitation of growth and reduction of seedling weight and seedling dried weight under stress. Kinetin increases amylases activity in seeds of plants under stress (Stavir *et al.*, 1998). Also, these researchers found that the harmful effects of stress on seedling weight and seedling dried weight and amylase activity are returned by adding kinetin growth regulator exogenous in culture of pea seeds. These substances neutralized stress conditions and by improvement of starch metabolism and amylase activity in cotyledon increased seedling weight and seedling dried weight growth. But in this present study highest seedling length was obtained in the control which was significantly higher than the next obtained in ethrel 1 ppm while the highest fresh weight was obtained in the ethrel 1ppm which was followed by significantly lower ethrel 10ppm. Here also the results are in contrast to the earlier similar studies. Highest dry weight of the seedling was obtained in kinetin50ppm treatment which was followed by kinetin 1ppm. The vigour index of the seedling was found to be the best in control condition which was followed by ethrel 1ppm. Thus this study showed that kinetin and also ethrel exhibit inhibitory effect on seed germination.

CONCLUSIONS

In conclusion, the present study showed that seed treatment with ethrel and kinetin exhibited the inhibitory effect on seed germination and seedling growth. None of the seed treatments provided superior germination over the control. Also, the seed treatments require additional steps and are more expensive. Only in case of seedling fresh and dry weight kinetin and ethrel showed slight superiority over the control. So, further studies are needed to elucidate whether these two growth regulators under specific light and thermal conditions have synergistic or additive effects on the germination of cucumber seeds and their subsequent seedling growth.

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APPENDICES

Table 1: Different Concentrations of Kinetin and Ethrel used in the Experiment

Treatments	Plant hormone used	Concentration of hormone solutions
S1	Kinetin	1 ppm
S2	Kinetin	10 ppm
S3	Kinetin	50 ppm
S4	Kinetin	100 ppm
S5	Ethrel	1 ppm
S6	Ethrel	10 ppm
S7	Ethrel	50 ppm
S8	Ethrel	100 ppm
Control	Distilled water	

Table 2: Effects of Different Treatments on Seed Germination and Seedling Growth of Cucumber

Treatment	Germination Percentage (%)	Germination Index (GI)	Radical Length (cm)	Plumule Length (cm)	Seedling Length (cm)	Fresh Weight (g)		Seedling Fresh wt(g)	Dry weight (g)		seedling Dry wt(g)	Vigour Index
						Radical	Plumule		Radical	Plumule		
S1	100 (90.00) ^a	7.4508 ^{de}	2.1800 ^{cd}	3.9500 ^b	6.1300 ^c	0.3770 ^d	1.3850 ^d	1.7620 ^a	0.0181 ^d	0.1260 ^b	0.1441 ^b	613.00 ^c
S2	70 (56.998) ^a	5.9000 ^e	0.971 ^f	2.7143 ^c	3.6857 ^{de}	0.1930 ^f	1.1390 ^f	1.3320 ^b	0.0098 ^f	0.0791 ^f	0.0889 ^g	258.00 ^d
S3	100 (90.00) ^a	8.1786 ^d	2.422 ^c	3.6500 ^b	6.0700 ^c	0.2210 ^e	1.8210 ^c	2.0420 ^d	0.0146 ^e	0.1460 ^a	0.1606 ^a	607.00 ^c
S4	80 (63.93) ^a	8.0206 ^d	0.537 ^f	2.6125 ^c	3.1500	0.1390 ^f	1.3230 ^e	1.4620 ^g	0.0074 ^g	0.0890 ^e	0.0964 ^f	252.00 ^d
S5	90 (75.00) ^a	19.2524 ^c	4.45 ^b	6.6111 ^a	11.0556 ^b	1.0160 ^b	2.1760 ^a	3.1920 ^a	0.0189 ^c	0.0723 ^g	0.0912 ^g	995.00 ^b
S6	100 (90.00) ^a	21.2619 ^b	1.900 ^{de}	2.8500 ^c	4.7500 ^d	1.2050 ^a	1.7910 ^c	2.9960 ^b	0.0295 ^a	0.0993 ^d	0.1288 ^d	475.00 ^c
S7	100 (90.00) ^a	23.7063 ^a	1.090 ^{ef}	1.8500 ^d	2.9400 ^{ef}	0.3690 ^d	1.1510 ^f	1.5200 ^f	0.0269 ^b	0.1114 ^c	0.1383 ^c	294.00 ^d
S8	100 (90.00) ^a	23.1508 ^a	0.850 ^f	1.6900 ^d	2.5400 ^f	0.2220 ^e	1.1240 ^f	1.3460 ^b	0.0151 ^e	0.1224 ^b	0.1375 ^c	254.00 ^d
Control	100 (90.00) ^a	24.9286 ^a	6.6 ^a	6.3200 ^a	12.9200 ^a	0.8300 ^c	2.0960 ^b	2.9260 ^c	0.0184 ^d	0.1009 ^c	0.1193 ^a	1292.00 ^a
Sed	4.556	.7897	.355	.246	.474	.0062	.0191	.0195	.0002	.0016	.0017	61.367
CD	11.435	1.9822	.891	.617	1.189	.0155	.0479	.0490	.0005	.0041	.0044	154.032

For statistical analysis, the data of germinating percentage was transformed to $\arcsin \sqrt{100/X}$. values in the parenthesis are arcsine transformed.

* Figures in the same column not sharing the same letters differ significantly at $p < 0.005$